GREENBUG (*SCHIZAPHIS GRAMINUM* RONDANI) BIOTYPE E INTERACTION WITH SORGHUM (*SORGHUM BICOLOR* L.) HYBRIDS.

by

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PART I

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INTRODUCTION

The greenbug, (Schizaphis graminum (Rondani) (Homoptera: Aphididae), is a cosmopolitan aphid with a host range of at least 60 species of grasses including most of the cereals (Walgenbach et al., 1988). In the midwestern United States, this insect is a major pest of wheat, Triticum aestivum L., barley, Hordeum vulgare L. and sorghum, Sorghum bicolor (L.) Moench, (Starks and Burton, 1977). Greenbugs have been a serious pest of wheat in the United States since 1882 and of sorghum since 1968 (Kindler et al., 1984). Greenbugs can cause substantial yield losses in crops by the direct effects of feeding or as vectors of several plant viruses (Walgenbach, et al., 1988). Along with other species of aphids greenbugs may transmit the causal agent of plant diseases, such as yellow dwarf and maize dwarf mosaic (Starks and Burton, 1977).

Greenbugs reproduce rapidly and mainly, in not entirely, by parthenogenesis (Mayo and Starks, 1972). When extremely abundant, greenbugs cause reduction in root and leaf development. In addition the number of tillers is usually reduced and the plant may be killed (Higgins and Brooks, 1987).

Five major greenbug biotypes (A, B, C, D and E) have been reported in the Great Plains of the United States (Starks et al., 1983). Greenbug biotype A (GBA) was followed by biotype B (GBB) which is virulent to wheat (Triticum aestivum L.) cultivar Dickinson Selection 28-A wheat while GBA is not (Starks et al., 1983). Biotype B was dominant during the early 1960's but was replaced by biotype C (Harvey and Hackerott, 1969) The appearance of biotype C (GBC) resulted in a severe outbreak of greenbugs on grain sorghum in 1968. This biotype was able to withstand

summer temperatures, and male alates were prevalent during certain times of the year (Mayo and Starks, 1974). Biotype B and C can be separated by their reaction to 'Piper' sudangrass, Sorghum sudanese (Peper) Starf, in the seedling stage. Piper is highly resistant to biotype B but susceptible to biotype C (Harvey and Hackerott, 1969). Biotype D (GBD) occurred and was resistant to organophosphate insecticides (Teetes et al., 1975). In 1980 a greenbug biotype, E (GBE), appeared and was able to attack GBC-resistant wheat (variety Amigo) according to Porter et al., (1982). Recently, other greenbug isolates have been discovered. These isolates include Ohio isolate (OH), designated as biotype F, Maryland isolate (MD), SCO isolate, designated as biotype G, and WCT isolate, designated as biotype H (Niemczyk 1980, Ratcliff and Murray 1983, Puterka et al., 1988). The Ohio isolate (biotype F) generally caused more severe injury on susceptible plants than did the Maryland isolate form. The Maryland isolate developed well on Kentucky blue grass (Poa pratensis L.) and barley (Hordeum vulgare L) while the Ohio isolate developed well only on Kentucky blue grass (Ratcliff and Murray, 1983). The SCO isolate (biotype G) damaged all known sources of greenbug resistance in wheat, (Triticum aestivum L.), but did not damage "Wintermalt', a barley, (Hordeum vulgare L.). The WCT isolate (biotype H) caused host plant responses on wheat that were similar to those of biotype E. However, biotype H was avirulent on sorghum and severely damaged 'Post', a barley variety resistant to all previously described biotypes (Puterka et al., 1988). These repeated occurances of new greenbug biotypes have drawn the attention of many researchers and some research has been conducted on the feeding behavior of greenbugs using

electronic monitors (Campbell et al., 1982; Montllor et al., 1983). The use of an electronic feeding monitor adds a new dimention to research on interaction between greenbug biotypes and sorghum germplasm sources. It may also provide data on variation within insect populations and estimation of frequencies of new biotype arising to overcome specific resistance mechanisms in specific germplasm sources (Bramel-Cox et al., 1986). In the feeding monitoring system, greenbug feeding activities are correlated with waveform patterns recorded in a millivolt strip-chart recorder (McLean and Kinsey, 1964). The feeding activities of aphids were initially identified and standardized by McLean and Kinsey (1964). Campbell et al., (1982) interpreted feeding monitor waveforms for sorghum and biotype C. They showed that feeding behavior of aphids is related to the resistance and susceptibility of the plant.

Chemical control is the main greenbug control when an outbreak occurs. High rates of persistant systemic insecticides were initially relied on to control greenbugs in sorghum. These treatments were effective but at the same time had broad-spectrum toxicity and were environmentally disruptive (Young and Teetes, 1977). Therefore, alternative approaches affording more economical control with less environmental contamination were sought. One such approach was greenbug resistance in sorghum (Harvey and Hackerott, 1969 and Wood, 1971).

Sources of resistant germplasm have now been located in all crop species (Starks and Burton, 1977) and because of this, many new sorghum hybrids have been released. Some of these hybrids have been studied extensively against GBE in terms of resistance and susceptibility in mature plants. However, very limited work has been done on these hybrids

against GBE in the seedling stage. Therefore, the first section of this research dealt with laboratory evaluation of sorghum hybrids against GBE in the seedling stage and assessed differences between resistant and susceptible sorghum in nearly isogenic sorghum hybrid pairs utilizing the following objectives:

1. Determine levels or magnitudes of resistance of seedling sorghum hybrids to GBE.
2. Evaluate which components or mechanisms of resistance are displayed by seedling sorghum hybrids:
   a. Antibiosis
   b. Antixenosis
   c. Tolerance
6. Describe feeding behavior of GBE on resistant and susceptible sorghum hybrids within a nearly isogenic pair in the seedling stage by the use of a feeding monitor.

The second section dealt with field evaluation of the advance growth stage sorghum hybrids relative to greenbug biotype E utilizing the following objective:

Determine the response and effect of greenbug biotype E on the advance growth stage sorghum hybrids in the field.

LITERATURE REVIEW

Levels or Mechanisms of Resistance

In 1983 Starks et al., reported that the level of resistance varied considerably among sorghum cultivars and that different cultivars had different mechanisms of resistance. Seedling resistance in sorghum is simply inherited and dominant, but the level of resistance was less in the heterozygote than in the homozygote (Hackerottet al., 1969; Weibel et al., 1972; and Teetes et al., 1974). Harvey and Hackerott, (1974) observed that susceptible sorghum seedlings infested with greenbugs were severely damaged and thus, less capable of supporting greenbug populations than resistant sorghum seedlings. In their study Weibel et al., (1972) observed that many dead or severely damaged sorghum seedlings were evident in susceptible rows and that there was migration of greenbugs from susceptible to resistant sorghum, 'Shallu Grain' (SA 7536-1), PI 264453 and IS 809 which were still green. Thus, aphid densities ranged higher on resistant entries and lower on susceptible ones. Similarly, they indicated that the average number of dead plants per entry and average reduction in height due to greenbugs were highly correlated to damage scores. Therefore, damage score is a good measure of resistance.

Components or Mechanisms of Resistance

Three mechanisms of resistance to insects were proposed by Painter (1951): (a) antibiosis or adverse effects of the host on the biology of insects; (b) antixenosis or behavioral term describing the ability of an individual insect to make active choices among a range of plants and (c)

tolerance or ability of a host plant to resist injury while supporting insect populations capable of damaging a susceptible host. The reduced survival and reproduction of an adult greenbug on resistant seedlings suggests antibiosis and antixenosis. Hackerott et al., (1969) reported that KS 30, PI 30108 and TS 1636 were resistant to greenbug biotype C. When preference, longevity and fecundity of greenbug biotypes (A, B & C) were studied on resistant SA 7536-1, KS 30, PI 264453 and susceptible RS-610 sorghums, they showed marked differences in development, i.e., biotype A did not survive on resistant species except to a limited degree on SA 7536-1 and KS 30. Biotype B performed slightly better than A on resistant species and much better than A on RS-610. Biotype C survived and reproduced on the resistant species almost as well as on RS-610 (Wood, 1971). Teetes et al., (1974) indicated that biotype C greenbugs displayed differential preference reactions when given a choice of biotype C-resistant or C-susceptible sorghum. They showed that resistant lines PI 264453, IS 809, KS 30 and 7536-1 were less preferred than susceptible sorghum TX 2536 and TX 7000. Teetes et al., (1974) studied the effects of susceptible and resistant sorghum on fecundity and longevity of greenbugs to evaluate antibiosis. Based on duration of nymphal stadia, they indicated that greenbugs reared on resistant sorghum SA 7536-1 and KS 30 required longer development periods than when reared on PI 264453 or susceptible TX 7000 and 2536 sorghum. The prereproductive period was longer and the reproductive period reduced for greenbugs reared on resistant versus susceptible sorghum. Greenbugs reared on SA 7536-1, KS 30 or IS 809 produced fewer progeny than those

reared on PI 264453, TX 7000 or TX 2536. Adult greenbugs lived longer on TX 7000 and TX 2536 than on SA 7536-1 or IS 809. Wood (1971) reported resistance in these sorghum lines: PI 264453, PI 220248, PI 308976, PI 302178, PI 302231 and SA 7536-1 and concluded that all three mechanisms of resistance were present. Resistant sorghum hybrids displayed varying degrees of resistance to greenbug feeding in the seedling stage. Preference tests indicated that hybrids E-59+ and E-59++ were more preferred than E-59 for the growth stage tested (Morgan, 1978). Boozaya-Angoon (1983) studied components of resistance to GBE on sorghum. She showed that PI 220248 and PI 264453 demonstrated high levels of antibiosis, non-preference and tolerance. Similarly, Schuster and Starks (1973) indicated that tolerance may be the main component of resistance in PI 264453. PI 229828, IS 809, Shallu grain, PI 302178 and PI 226096 displayed comparatively high degrees of all resistance components. Morgan (1978) indicated that the overall superior tolerance of two parent resistant hybrid E-59++ suggests that homozygous resistant hybrids are advantageous in the seedling stage. Similarly, the homozygous resistant line (H-39) and heterozygous F_1 appeared to be resistant enough to tolerate a seedling infestation without sustaining significant yield losses (Harvey and Hackerott, 1974). Bramel-Cox _et al_., (1986) found the best new source of overall resistant to be PI 266965 while the best sources of tolerance, antibiosis and antixenosis were PI 229828, PI 266965 and J 242, respectively.

Electronic Monitoring of Greenbug Feeding

The use of an electronic feeding monitor adds a new dimension to research on interaction between greenbug biotypes and sorghum germplasm sources. It may also provide more data on variation within insect populations and estimation of frequencies of new biotypes arising to overcome specific resistance mechanisms in specific germplasm sources. The mechanism of resistance measured by a feeding monitor probably relates most closely with antibiosis, but the evidence is still preliminary (Bramel-Cox, et al., 1986). A technique was developed when aphids salivate and ingest within an electronically conductive substance (McLean and Kinsey, 1964). These activities were identified and standardized by correlating them with specific curve patterns recorded in a millivolt strip-chart recorder (McLean and Kinsey, 1965). Since the development of this device modifications have been made as described by McLean Kinsey, (1967); McLean and Weight, (1968); and Brown and Holbrook, (1976). In the feeding monitoring system, the aphid is connected to the electrical circuit with a fine gold wire (10 microns) attached to its dorsum. When the aphid starts to probe the leaf, the circuit becomes complete and the chart recorder records different wave forms corresponding to the different feeding activities (McLean and Kinsey, 1967; Brown and Holbrook, 1976). There are five distinct wave forms identified corresponding to the feeding activities: (1) probing (2) salivation (3) Non-phloem ingestion (4) stylet penetration of sieve elements and (5) phloem ingestion by the aphid (McLean and Kinsey, 1967; and Campbell et al., 1982).

Probing is the first physical contact of the aphid's stylets to the host plant. Aphids make test probes prior to ingestion and an increased number of separate probes or an increased duration of non-probing were associated with resistant lines (Campbell et al., 1982). Salivation occurs during probing and is characterized by the formation of sheath material from the time of initial probing to the location of vascular bundles. The total salivation time of feeding on resistant varieties is longer compared to feeding on susceptible varieties (Nielson and Don, 1974). However, according to Campbell et al., (1982), there was no significant difference in mean duration of salivation between resistant and susceptible varieties. Aphids sometimes feed on non-phloem tissues, such as mesophyll and parenchyma cells. According to Campbell et al., (1982) differences in mean duration of non-phloem ingestion by greenbugs were not definitively correlated to resistance in sorghum. Correlation between lack of, or reduced ingestion from the phloem and aphid probing resistant and non-host plants have been reported from other crop species (Kennedy et al., 1978; McLean and Kinsey, 1968; Nault and Styer, 1972; Campbell et al., 1982).

A combination of salivation waveforms, x-wave forms and ingestion wave forms are usually observed when aphids feed (McLean and Weight, 1968; Nielson and Don, 1974; and Campbell et al., 1982). X-wave forms are produced when the stylet penetrates the sieve elements in the phloem and they always precede ingestion wave forms (Campbellet al., 1982; McLean and Kinsey, 1967). The ingestion wave forms indicate withdrawal of the sap from the sieve element. The duration of phloem ingestion is longer on susceptible than on resistant hosts (Campbell et al., 1982;

and Montllor et al., 1983). The differences in chemical constituents of the phloem between susceptible and resistant varieties might account for the differential feeding behavior of greenbugs (Campbell et al., 1982; Nielson and Don, 1974; and Kennedy et al., 1978). McLean and Kinsey (1968) compared salivation and ingestion from host and non-host leaves. They found that significant differences in probing behavior occurred between aphids Acyrthosiphum pisum [Harris] on host and non-host plants. In the same study they observed statistical differences in probing behavior between aphids on healthy Vicia faba and those on diseased Vicia faba. Studies by Campbell et al., (1982) on probing behavior of biotype C greenbug on susceptible and resistant sorghum lines showed that aphids probing the resistant lines significantly reduced imbibition of phloem sap compared with aphids fed on susceptible varieties. Also, increased numbers of probes and increased duration of non-probing were associated with greenbug feeding on resistant lines (Campbell et al., 1982). Biotype E greenbugs were found to grow and reproduce at approximately twice the rate of biotype C on biotype C-resistant sorghum IS 809 (Montllor et al., 1983). When probing behavior of both biotypes was electronically monitored on IS 809, biotype E time to first committed phloem ingestion (i.e. ingestion from the phloem lasting > 15 minutes) was significantly shorter than that for greenbug biotype C. Similarly, the total duration of phloem ingestion during a 24 hour period was significantly longer for GBE than for GBC, but this would be accounted for by the shorter time needed for aphids of GBE to establish initial CPI (Montllor et al., 1983). Further experiments demonstrated that GBC exposed to IS 809 for at least 24 hours prior to being

monitored on this variety also reached the phloem faster, time to CPI was shorter and they spent a longer time feeding from the phloem than did biotype C greenbugs without previous exposure to this variety (Montllor et al., 1983).

Effect of Advance Growth Stage Sorghum Hybrids Against GBE in the Field

Johnson et al., (1974) observed that, in untreated plots, the mean number of greenbugs (GBC) per plant was higher and leaf injury from greenbug feeding was greater in susceptible entries than in resistant lines and their hybrids in the field on mature sorghum plants. Similarly, they observed that hybrids with one resistant parent exhibited enough resistance to control greenbug populations and significantly reduce leaf death from greenbug feeding on adult plants. Agronomically improved greenbug resistant sorghum hybrids infested (naturally) with fewer greenbugs were damaged less and produced higher yields than genetically comparable greenbug susceptible hybrids under natural insect infestations (Teetes et al., 1975). Sorghum lines and F_1 hybrids were screened in the field in mature plants against GBC and the results indicated that sorghum lines IS 809, KS 30 and SA 7536-1 in general, were significantly less infested with greenbugs than susceptible lines TX 7000 and TX 2536. F_1 hybrids displayed similar results when compared with susceptible NB 505 (Teetes et al., 1974).

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PART I

LABORATORY EVALUATION OF SEEDLING SORGHUM HYBRID RESISTANCE TO GREENBUG BIOTYPE E.

Abstract

The effect of varying greenbug infestation levels, identification of components of resistance and feeding behavior of greenbug biotype E were studied on seedling sorghum in the laboratory.

Increased greenbug infestation levels affected sorghum seedlings by reducing leaf area and hence, plant dry weight. However, the effect was more on the susceptible than the resistant hybrid. The resistant members of nearly isogenic hybrid pairs which demontrated relatively high levels of resistance include E 1616(R)/HW 7217(S) and G 550E(R)/HW 2194(S). Antibiosis and or antixenosis were the main components of resistance in these hybrids. The highest antibiosis and antixenosis levels were found in the resistant member of hybrid pair 758(R)/6073(S) and high levels of tolerance were observed in the resistant member of hybrid pair 728(R)/DK 28(S). The feeding behavior studies indicated that greenbug biotype E spent more time feeding on the susceptible hybrid 6073 than it did on the resistant member 758.

INTRODUCTION

Greenbug, Schizaphis graminum (Rondani) (Homoptera: Aphididae), is a cosmopolitan pest of many graminaceous crops. The greenbug has been rated as a key pest in sorghum, based upon its perennial occurence and its ability to consistently exceed economic thresholds (Young and Teetes, 1977). The insects remove photosynthates, inject toxins that kill leaves, are vectors of certain viruses and predispose plants to diseases such as charcoal rot (Young and Teetes, 1977).

Three mechanisms of resistance to insects were reported by Painter (1951). Antibiosis is the tendency of crops to prevent, injure, or destroy (insect) life. The effects on the insect take the form of reduced fecundity, decreased size, abnormal length of life and increased mortality. Antixenosis refers to the group of plant characters and insect responses that lead away from the use of a particular plant or variety for oviposition, for food, for shelter, or for any combination of the three. Tolerance is a basis of resistance in which the plant shows an ability to grow and reproduce itself or to repair injury to a marked degree in spite of supporting a population approximately equal to that damaging a susceptible host.

Resistant hybrids and varieties have been released to counteract the damaging activities of greenbugs. However, the levels of resistance vary considerably among cultivars and different cultivars may have different mechanisms of resistance (Starks et al., (1977).

Greenbugs feed by inserting their stylets and withdrawing sap from the plants. Salivation occurs as they dissolve their way enzymatically

down through the pectin to the phloem. From this feeding behavior, McLean and Kinsey, (1964) developed a technique to determine when aphids salivate and when they ingest within an electrically conductive substance. These feeding activities were then identified and standardized by correlating them with specific curve patterns, recorded in a millivolt strip-chart recorder (McLean and Kinsey, 1965). Since the development of this device, modifications have been made as described by McLean and Kinsey, (1967); McLean and Weight, (1968); Brown and Holbrook, (1976). The feeding behavior recorded in this device was related to resistance and susceptibility. From this relationship, McLean and Kinsey, (1968); Campbell et al., (1982); Nielson and Don, (1974); Montllor et al., (1983) demonstrated that greenbugs spent a longer time feeding on susceptible plants than those feeding on resistant plants.

The objectives of this research were (1) to measure levels of resistance and (2) identify mechanisms of resistance to greenbug biotype E (i.e. antibiosis, antixenosis and tolerance) in seedling nearly isogenic sorghum hybrid pairs. Finally, I wanted to study the feeding behavior of greenbug biotype E on a selected seedling nearly isogenic sorghum hybrid pair in the laboratory in order to explore the relationship between resistance and feeding behavior.

MATERIALS AND METHODS

Five pairs of nearly isogenic commercial sorghum hybrids (5 resistant and 5 susceptible to GBE) were tested. These hybrids were obtained from different seed companies and were released in 1986-87. These hybrid pairs are:

Resistant	Susceptible
1. 708	DK 18
2. 728	DK 28
3. 758	6073
4. E 1616	HW 7217
5. G 550E	HW 2194

Greenbug biotype E was taken from a colony kept by Dr. Tom Harvey at Hays, Kansas. It was maintained in the greenhouse at KSU and when periodically screened proved still to be GBE (Dr. Reese, personal communication). A susceptible hybrid, Funk G 550, was used as a host for maintaining the greenbug colony in a rearing room at $25^{o}C$ - $29^{o}C$ with ca. 70% relative humidity.

Levels or Magnitudes of Resistance of Seedling Sorghum Hybrids

Three seeds from each hybrid were planted in a plastic supercell container (diameter = 3.8cm; depth = 20cm) held in holding crates (length = 72cm; width = 36cm) containing "Sunshine Mix" a complete soil medium for optimum plant growth from Fisons Western Corporation (Canada). The soil was tamped within 3cm of the top of the container.

One watering from the top was made to induce germination and subsequent waterings were by capillarity through the bottom of the container immersed in water. After emergence, plants were thinned to one vigorous seedling per container. There were four infestation levels of adult greenbugs, 0, 5, 10 and 20/plant. When seedlings of each hybrid were at the three leaf stage, each was infested with one of the four levels of greenbugs, therefore this was a 10x4 factorial experiment. This experiment was conducted twice in a randomized complete block design with 6 replications. After infesting with greenbugs, a clear cage of 2.5 x 30 cm with 3 air holes (about 15 cm in diameter) covered with a fine sieve cloth was placed on each container to prevent greenbugs from escaping. The growth chamber environment was a 16L:8D photoperiod at 27 $\pm$ 2 ^{o}C. One week post-infestation live greenbugs on each seedling were counted. All sorghum seedlings were rated for greenbug damage, using a scale of 0 (no damage) to 10 (death of plant). Each leaf was rated and the mean damage for the whole plant obtained. Leaf area for each leaf of each entry, as measured by a Licor^{R} area meter with belt attachment, was recorded. To determine the functional leaf area, the damage rating for each leaf in proportion damaged was multiplied by the leaf area of that leaf and subtracted from 1. Total functional leaf area for a plant was obtained by adding the functional leaf area of all leaves on the plant. Each seedling was oven dried at 60^{o}C for 48 hours and placed in a dessicator. Weight was recorded after the sample had reached a constant weight. All agronomic characteristics tested were subtracted from their respective noninfested check and then divided by the noninfested check to obtain the proportion reduction. The proportions were subsequently

multiplied by one hundred to obtain percent reductions as shown by the equation below:

$$\text{Percent Reduction} = \left(\left(\frac{\text{Noninfested-Infested}}{\text{Noninfested}}\right)\right) \times 100$$

Means for damage ratings and agronomic characteristics i.e. functional leaf area reduction (%) and plant dry weight reduction (%) were analyzed for each infestation level for each hybrid by using GLM. Regression analysis on the raw data for each parameter versus greenbug infestation levels was performed in order to determine if an increase in greenbug infestation levels would affect the agronomic characteristics tested. Furthermore, a paired t-test analysis was performed for each isogenic pair at each infestation level for the parameters tested. Finally, correlation coefficients were calculated to determine the effect of increasing greenbug numbers on the parameters tested.

Components or Mechanisms of resistance

Two methods were used to measure the antibiotic effect of sorghum hybrid seedlings against GBE. The first method measured the development period (days) and fecundity per individual greenbug while the second method measured only fecundity per five greenbugs on the nearly isogenic sorghum hybrid seedlings.

A. Antibiosis test, part 1.

Planting method and experimental conditions were the same as in the preceeding experiment, except greenbug levels were constant. Each seedling was infested with three adult greenbugs. At 24 hours post

infestation, all greenbugs except one neonate per seedling were removed. The seedlings were observed daily as the neonate developed. When it became a reproductive adult, the nymphs produced were counted and removed until the female greenbug ceased nymph production. This test determines the variation in fecundity, rate of development and length of life span of females reared on susceptible and resistant sorghum hybrids. The experiment was conducted twice using a randomized complete block design with six replications. A paired t-test and analysis of variance procedure were used to analyze the data within nearly isogenic pairs. Mean separation by LSD for the resistant versus resistant and susceptible versus susceptible was performed to determine which hybrid had the most and the least antibiotic effect on the greenbug (GBE).

Antibiosis test part 2.

The planting procedure was the same as the antibiosis test part 1. However, the test consisted of infesting the seedling with five apterous adult greenbugs and, after five days, removing all but five nymphs uniform in size (Bramel-Cox _et al_., 1986). The nymphs were allowed to develop and new adults reproduce on the seedlings for eight days after which all greenbugs were removed and counted. Paired t-test, analysis of variance and mean separation by the least significant difference (LSD) procedures were performed as discussed above.

B. Antixenosis

Experimental conditions were the same as the preceeding experiments. One nearly isogenic hybrid pair (resistant and susceptible

pairing as shown below) was planted in one container (ca. 9 cm diameter). Three seeds of each hybrid were planted per hole in one container opposite from each other. After plant emergence, seedlings were thinned to one uniform vigorous seedling of each hybrid. When seedlings were at the 3 leaf stage, 25 greenbug nymphs (3-4th instar) were placed at the center of the container. The container was covered with a cage to prevent greenbugs from escaping. Greenbug counts on each seedling were taken 48 hours post infestation. The experiment was repeated three times in a randomized complete block design with six replications. Greenbug counts were converted into proportions of the initial number of greenbugs. Due to the fact that the number of greenbugs counted on both plants did not equal 25, the original number infested, and the variability of the data, an arcsine transformation was utilized on the proportions and a paired t-test was used to compare the counts in each nearly isogenic hybrid pair. Nearly isogenic hybrids were paired as follows:

Resistant	Susceptible
1. 708	DK 18
2. 728	DK 28
3. 758	6073
4. E 1616	HW 7217
5. G 550E	HW 2194

C. Tolerance test.

Each hybrid was planted in two containers i.e. infested and noninfested treatments. After plant emergence, containers were thinned

to one vigorous seedling per container. When plants reached the three leaf stage, plant height of both infested and noninfested treatments was determined from the soil level to the tip of the longest leaf. One of the two containers was infested with five apterous adult greenbugs while the other was a noninfested control. Eight days post infestation, greenbug counts were made and plant heights of all entries were re-determined. Similarly, the above-ground dry weight of each hybrid in an infested and noninfested plant was determined. This experiment measures how greenbugs can affect both plant height and dry weight of resistant and susceptible sorghum in the seedling stage. The experiment was conducted twice in a randomized complete block design and each experiment had six replications. Tolerance of a hybrid was based upon plant indices, which were adjusted for differences in levels of antibiosis. This technique estimates tolerance according to its theoretical definition much better than a visual rating scale (Bramel-Cox *et al*., 1986). The plant indices were calculated as follows:

$$\text{Plant Index} = \frac{\left(\frac{(\text{Noninfested}-\text{Infested})}{\text{Noninfested}} \times 100\right)}{\text{Greenbug number}}$$

A paired t-test was used to compare tolerance within a pair of each nearly isogenic pair. Analysis of variance procedure and mean separation by least significant difference were done on the indices obtained.

Electronic Monitoring of Greenbug Feeding

Electronic feeding monitors which had been modified several times from the original equipment described by Mclean and Kinsey (1964, 1967) were used. Each monitor utilized in this experiment had a 9V battery. Included was a 25-Hz oscillator providing 200mV AC current to the soil of the test plant. To prepare the aphid for monitoring, a drop of silver glue was placed on a petri dish and the tip of the 10 micron gold wire (ca. 4cm long) was dipped in the drop until a small ball was formed. The greenbug dorsum was pressed to the moist ball of silver glue and thereby attached to a wire which was connected to the monitor. One pair of nearly isogenic sorghum hybrids (758(R)/6073(S)) was used and ten GBE were monitored for each hybrid on ten separate plants. This nearly isogenic pair was selected based on the relatively high antibiosis of its resistant member against greenbugs (GBE) observed in the preliminary studies. Each greenbug was placed on the adaxial surface of the last fully expanded leaf. When the greenbug started to feed, an electric circuit was completed and the signal was amplified and rectified before being transmitted to the computer. Monitoring was accomplished uninterruptedly for 12 hours for each run. Any greenbugs that had fallen off the plant during the course of the experiment were not replaced and therefore not included in the analysis. Individual records of ten greebugs were obtained and analyzed.

The following parameters were considered:

A. Total number of separate probes (i.e. stylet removed and re-entered in plant tissue during a period of twelve hours).

B. Time to first committed phloem ingestion (CPI) measured as minutes to the first x-wave followed by > 15 minutes of ingestion from the phloem.

C. Total frequencies of phloem ingestion (Feeding).

D. Total phloem ingestion (Feeding time in minutes).

E. Total number of x-waves produced during a period of twelve hours.

F. Total salivation.

G. Total baseline.

The data were analyzed by the paired t-test and correlation among the parameters was performed.

RESULTS AND DISCUSSION

Levels or Magnitudes of Resistance

The effect of varying infestation densities of greenbugs (GBE) on selected agronomic characteristics of seedling sorghum were compared and statistical differences apparent. Each hybrid was analyzed separately for the parameters tested by the greenbug infestation levels and in all cases the greater the greenbug density the greater the effect on the plant (Tables 1-3). Thus, as might be expected, the greater number of greenbugs infesting seedling stage sorghum, the greater the plant is stressed causing a reduction in functional leaf area, and dry weight which, if the plants survived, would probably have a negative impact upon yield.

To measure the magnitude of the effect of greenbugs on the plant characteristics and damage, regression analysis was performed. In general, hybrids responded similarly to increasing levels of greenbug numbers. This suggests that members of each isogenic pair were affected equally by an increase in the greenbug infestation levels tested. No significant differences were observed in terms of functional leaf area reduction (%) when the slopes of members of nearly isogenic sorghum hybrid pairs were compared. When slopes of damage ratings and plant dry weight were compared between the members of each nearly isogenic pair, statistical differences were evident in only one nearly isogenic pair 728(R)/DK 28(S). The susceptible member had a higher slope than the resistant counterpart (Table 4). This indicates that the susceptible member in this nearly isogenic pair was more sensitive to increasing

greenbug numbers than the resistant hybrid.

Furthermore, to determine the effect of greenbugs on the nearly isogenic pairs, a paired t-test analysis on the plant characteristics and damage at different greenbug infestation levels was performed. In general, greenbugs caused higher leaf area reduction (%) on susceptible than on resistant members in each pair (Table 5). However, functional leaf area reduction (%) differed significantly only at the higher levels of greenbug infestation tested. The resistant member of the nearly isogenic hybrid pairs which differed significantly in terms of functional leaf area reduction included 758, E 1616 and G 550E. Similarly, damage ratings differed significantly between the members of hybrid pairs 758(R)/6073(S) and E 1616(R)/HW 7217(S) at all greenbug infestation levels tested, while a significant difference was observed only when the initial infestation level was 10 greenbugs in hybrid pair G 550E(R)/HW 2194(S) (Table 6). In each case the susceptible member in the pair had more necrotic leaves than the resistant counterpart. When comparing nearly isogenic pairs, plant dry weight reduction (%) was not significantly different at practically all greenbug infestation levels tested.

Pearson correlation coefficients were calculated for each parameter with the results shown in Tables 7 and 8. As one might expect, percent leaf area reduction and percent plant dry weight reduction were positively correlated with both greenbug infestation levels and damage ratings. When greenbug infestation levels were increased, percent leaf area reduction tended to increase due to the damage resulting from greenbug feeding. Reduction in leaf area affected plant photosynthesis

and as a result percent plant dry weight reduction was also increased.

Components or Mechanisms of Resistance

Antibiosis tests

Table 9 presents results of the effect different hybrids have on greenbug (GBE) life stages in days for each pair of nearly isogenic sorghum hybrids. Also included in this table are the number of nymphs produced per female (fecundity) for each hybrid.

To compare resistant versus resistant and susceptible versus susceptible hybrids to determine which had the greatest and/or least antibiotic effect, the data were analyzed accordingly and is presented in Table 10. As illustrated in both Tables 9 and 10 the hybrid having an overall greater antibiotic effect upon GBE is hybrid 758(R). Greenbugs feeding on this hybrid had the shortest post-reproductive life span and the shortest total life span which resulted in the least number of nymphs produced per female of any of the resistant hybrids. It was also the only nearly isogenic hybrid that differed significantly in all three of these parameters from it's nearly isogenic partner. This is an important effect because by reducing both duration of life span and number of young produced the probability of reducing the amount of damage to the plant is increased especially to seedling stage sorghum. None of the hybrids significantly affected nymphal duration or reproductive life span. Other parameters did not differ significantly in some of the nearly isogenic pairs. The results of the second antibiosis test are presented in Tables 11 and 12 and support those obtained from the trial relative to greenbug numbers. Hybrid 758(R) still seemed to

exhibit the greatest effect upon the greenbugs in reducing fecundity. Therefore this hybrid seems to have a deleterious effect on length of life thereby having the potential of less feeding and at the same time it seems to cause fewer offspring to be produced, thus further reducing the damage potential. Nearly isogenic hybrid pairs 708(R)/DK 18(S), 728(R)/DK 28(S) did not differ significantly in greenbug numbers between the nearly isogenic members even when more than one greenbug were reared per entry. Hybrid pair G 550E(R)/HW 2194(S) revealed significant differences in greenbug numbers between its nearly isogenic members when more than one greenbug was utilized in this experiment. Therefore, the non-significant differences obtained in the antibiosis part 1 may be related to the single greenbug per plant utilized in that experiment.

Antixenosis

Trials were conducted to compare any antixenotic effect which may be exhibited by the nearly isogenic sorghum hybrids to greenbug biotype E. These results are presented in Table 13. As with the antibiosis trial, hybrid 758(R) seemed to exhibit the greatest antixenotic effect. Resistant hybrid E 1616(R) differed signicantly from its nearly isogenic partner. As measured by the previous antibiosis test this hybrid, E 1616(R), had a somewhat deleterious effect upon GBE; thus, it appears to have both mechanisms, antibiosis and antixenosis, also and was the only other hybrid, besides 758(R), to differ significantly from its nearly isogenic partner.

Tolerance test

To measure the seedling sorghum hybrid's relative ability to

withstand greenbug infestations, i.e. tolerance, agronomic characteristics of plant height and plant dry weight were compared as an index of greenbug effect between the nearly isogenic hybrid pairs (Table 14). Similarly, comparisons between the resistant and susceptible hybrids were performed (Table 15). Since this index compares the relative percent plant height and dry weight reduction per greenbug it does away with the antibiotic effect the hybrids may have upon the greenbug and therefore seems to be a true measure of the plant's ability to tolerate biotype E greenbugs. As illustrated in Table 14 greenbugs had the least effect on hybrid 728(R) in reducing plant height and there was no significanct difference in dry weight reduction between this hybrid and 758(R) which had the least reduction in dry weight. Therefore, as far as the plant's ability to tolerate greenbug infestations is concerned it seems that hybrid 728(R) would have to be classified as overall, the most tolerant. It's nearly isogenic member (DK 28) was also relatively tolerant when compared to the other susceptible hybrids. Although there were no significant differences in plant height reduction among the susceptibles, hybrid DK 28(S) ranked the second most tolerant as it also did for dry weight reduction. The least tolerant hybrid, E 1616(R), was affected by greenbugs (GBE) the most in both categories measured but its nearly isogenic member was the least affected, as far as plant height was concerned, of any of the susceptibles. Hybrid 758(R) which seemed to be the resistant hybrid displaying the greatest antibiotic and antixenotic effect to greenbugs (GBE) did not differ significantly from 728(R) in plant dry weight reduction but was significantly different for the characteristic of

plant height. This characteristic, height, may not be as important, ultimately, as it relates to standability or yield as overall dry weight. A plant may be reduced in height because of insect feeding, however, the component most important to sorghum producers, seed production, may not be reduced as significantly if the plants' overall leaf and stem tissues are not as affected and are able therefore to be utilized for seed production. From the standpoint of all three resistance mechanisms in the seedling stage, hybrid 758 seems to have the most potential relative to the resistant hybrids included in this study.

Electronic monitoring of greenbug feeding

Feeding behavior of GBE was studied for a twelve hour period on one pair of nearly isogenic sorghum hybrids 758(R)/6073(S).

Feeding time was the only parameter which differed significantly ($P \leq 0.05$) between the resistant and the susceptible members of the hybrid pair 758(R)/6073(S). Greenbugs spent less time feeding on the resistant hybrid 758 than those feeding on the susceptible member 6073 (325.41min. versus 521.63min., respectively). Number of separate probes, mean frequencies of salivation, x-waves, phloem ingestion, non-phloem ingestion and time to first committed phloem ingestion did not differ significantly between the two members of the pair tested. These non-significant differences may be brought about by the 'tethering effect' i.e. the attachment of the gold wire to the greenbug. Wiring reduced the quantitative differences between the aphid reactions to host and non-host plants significantly in a previous study by Tjallingi (1978).

Pearson correlation coefficients were calculated and results indicate feeding time to be negatively correlated with fequencies of baseline, probing and salivation at $P \leq 0.05$. Thus, the more time greenbugs spend in performing activities other than feeding the greater the negative correlation, i.e. the less time they actually have available for feeding.

Less time spent in feeding on the resistant hybrid could result in an antibiosis and or antixenosis mechanism exhibited by this hybrid. These results support earlier results obtained in the antibiosis and antixenosis experiment discussed above.

SUMMARY

In general, increasing levels of greenbug infestation increased damage equally among the hybrids. However, various degrees of resistance were exhibited by the hybrids tested. Resistant hybrid 758, E 1616 and G 550E manifested some level of antibiosis and antixenosis effect on greenbug biotype E but the other two tested (708 and 728) did not. Starks et al., (1983) reported that levels of resistant varied considerably among cultivars and different cultivars had different mechanisms of resistance. Teetes et al., (1974), Hackerott et al., (1969), Wood (1971), Bramel-Cox et al., (1986) and Boozaya-Angoon, (1983) reported that greenbugs reared on susceptible cultivars reproduced and lived longer than those greenbugs reared on resistant cultivars. Similarly, they indicated that resistant cultivars were less preferred than the susceptible cultivars. However, Morgan (1978) found no antixenosis in the seedling sorghum hybrids resistant to biotype C he

tested.

Only one of the resistant hybrids (728) exhibited a significant level of tolerance as measures by a plant height index and dry weight index in the seedling stage. Bramel-Cox _et al_., (1986) measured tolerance in terms of weight indices and indicated that the most tolerant line was PI 229828 and the least tolerant were Redlan and PI 302136.

In terms of feeding damage, resistant levels in commercial sorghum hybrids resistant to greenbug biotype E appeared similar to that reported by Morgan (1978) to seedling sorghum hybrids resistant to greenbug biotype C in the seedling stage.

Feeding monitor results suggest that reduced feeding time may account for the antibiosis and or antixenosis exhibited by resistant hybrid 758. Studies by Campbell _et al_., (1982) and Montllor _et al_., (1983) indicated the duration of phloem ingestion (feeding) by the greenbug is longer on susceptible than on resistant hosts. Dorschner (Anon. 1989) found that aphids on the susceptible hop plant spent 87 percent of their time actually ingesting phloem sap, compared to 40 and 58 percent for aphids on the two resistant plants. From this study, Dorschner (Anon. 1989) suggested that since survival and multiplication of aphid populations were directly related to how much time they spent feeding, resistant plants may partially starve aphids. The differences in chemical constituents of the phloem between resistant and susceptible varieties might account for the differential feeding behavior of greenbugs (Campbell _et al_., 1982, Nielson and Don (1974) and Kennedy _et al_., 1978).

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Table 1. Effect of greenbug infestation levels on functional leaf area reduction (%) for seedling sorghum hybrids 7 days postinfestation.[1]

Hybrids[2]		Infestation levels(GBE/plant) 5	10	20
708	R	17.65 c	42.77 b	81.70 a
728	R	26.53 c	54.84 b	78.05 a
758	R	18.09 c	41.37 b	71.49 a
E 1616	R	29.06 c	52.42 b	70.45 a
G 550E	R	18.19 c	43.61 b	63.36 a
DK 18	S	25.31 c	49.20 b	80.07 a
DK 28	S	36.41 c	69.20 b	84.31 a
6073	S	27.13 c	51.12 b	86.88 a
HW 7217	S	24.05 c	53.41 b	84.85 a
HW 2194	S	27.14 c	57.95 b	85.91 a

[1]Means with the same letter within horizontal rows are not significantly different at $P \leq 0.05$ by LSD.
[2]R and S represent resistant and susceptible nearly isogenic sorghum hybrids.

Table 2. Effect of greenbug infestation levels on dry weight reduction (%) for seedling sorghum hybrids 7 days postinfestation[1].

Hybrids[2]		Infestation levels(GBE/plant)		
		5	10	20
708	R	13.60 c	34.34 b	55.34 a
728	R	17.04 c	39.68 b	56.10 a
758	R	16.91 c	33.75 b	45.10 a
E 1616	R	24.70 c	41.60 b	54.71 a
G 550E	R	16.56 c	30.60 b	47.80 a
DK 18	S	21.76 c	37.42 b	60.07 a
DK 28	S	36.59 c	48.47 a	58.62 a
6073	S	19.72 c	37.43 b	53.39 a
HW 7217	S	14.94 c	32.11 b	57.41 a
HW 2194	S	18.16 c	38.64 b	60.99 a

[1]Means with the same letter within horizontal rows are not significantly different at $P \leq 0.05$ by LSD.
[2]R and S represent resistant and susceptible nearly isogenic sorghum hybrids.

Table 3. Damage ratings to seedling sorghum hybrids 7 days postinfestation resulting from differing greenbug infestation levels.[1]

Hybrids[2]		Infestation levels(GBE/plant)		
		5	10	20
708	R	2.40 c	3.58 b	5.94 a
728	R	2.25 c	3.65 b	5.31 a
758	R	2.33 c	3.21 b	5.13 a
E 1616	R	1.96 c	3.23 b	4.96 a
G 550E	R	1.94 c	2.92 b	4.98 a
DK 18	S	2.94 c	4.00 b	6.56 a
DK 28	S	2.94 c	4.67 b	6.06 a
6073	S	3.58 c	4.71 b	7.00 a
HW 7217	S	2.75 c	3.94 b	6.00 a
HW 2194	S	2.43 c	4.20 b	5.75 a

[1]Means with the same letter within horizontal rows are not significantly different at $P \leq 0.05$ by LSD.
[2]R and S represent resistant and susceptible nearly isogenic sorghum hybrids.

Table 4: Summary of regression analysis of damage ratings and dry weight versus greenbug infestation levels.

Hybrid pairs[2]		Slopes: Damage Ratings	Slopes: Dry Weight
708	R	0.21	-1.23
		ns	ns
DK 18	S	0.25	-0.94
728	R	0.21	-1.24
		*	*
DK 28	S	0.25	-1.34
758	R	0.19	-1.08
		ns	ns
6073	S	0.24	-1.34
E 1616	R	0.20	-1.14
		*	ns
HW 7217	S	0.22	-1.55
G 550E	R	0.21	-1.19
		ns	ns
HW 2194	S	0.21	-1.37

[1]R and S represent resistant and susceptible nearly isogenic sorghum hybrids respectively.
* and ns represent significance and nonsignificance at $P \leq 0.05$.

Table 5. Pairwise comparison of effect of greenbug infestation levels on functional leaf area reduction (%) of seedling nearly isogenic sorghum hybrids.[1]

Hybrid Pairs[2]		Greenbug Infestation Levels 5	10	20
708	R	17.65	42.77	81.70
		ns	ns	ns
DK 18	S	25.31	49.20	80.07
728	R	26.53	54.84	78.05
		ns	ns	ns
DK 28	S	36.41	69.20	84.31
758	R	18.09	41.37	71.49
		ns	ns	*
6073	S	27.13	51.12	86.88
E 1616	R	29.06	52.42	70.45
		ns	ns	*
HW 7217	S	24.05	53.41	84.85
G 550E	R	18.19	43.61	63.36
		ns	ns	*
HW 2194	S	27.14	57.95	85.91

[1]* and ns represent significance and nonsignificance at $P < 0.05$ by paired t-test.
[2]R and S represent resistant and susceptible nearly isogenic sorghum hybrids.

Table 6. Pairwise comparison of damage ratings to seedling sorghum hybrids resulting from differing greenbug infestation levels.[1]

Hybrid Pairs[2]	Greenbug Infestation Levels		
	5	10	20
708	2.40	3.58	5.94
	ns	ns	ns
DK 18	2.94	4.00	6.56
728	2.33	3.21	5.13
	ns	ns	ns
DK 28	2.94	4.67	6.06
758	2.33	3.21	5.13
	*	*	*
6073	3.58	4.71	7.00
E 1616	1.91	3.23	4.96
	*	*	*
HW 7217	2.75	3.94	6.00
G 550E	1.94	2.92	4.98
	ns	*	ns
HW 2194	2.43	4.20	5.75

[1]* ns represent significance and nonsignificance at $P < 0.05$ by paired t-test.
[2]R and S represent resistant and susceptible nearly isogenic sorghum hybrids.

Table 7. Pearson correlation coefficients (R) for greenbug infestation levels.

Hybrids[1]		LA[2]	DWT[2]
708	R	0.69 *	0.51 *
728	R	0.32 *	0.34 *
758	R	0.67 *	0.64 *
E 1616	R	0.70 *	0.69 *
G 550E	R	0.69 *	0.54 *
DK 18	S	0.50 *	0.47 *
DK 28	S	0.74 *	0.71 *
6073	S	0.33 *	0.50 *
HW 7217	S	0.52 *	0.61 *
HW 2194	S	0.66 *	0.64 *

[1]R and S represent resistant and susceptible nearly isogenic sorghum hybrids.
[2]LA and DWT represent percent leaf area and plant dry weight reductions.
* represent significance and nonsignificance at $P \leq 0.05$ by t-test.

Table 8. Pearson correlation coefficients (R) for damage ratings.

Hybrids[1]		LA[2]	DWT[2]
708	R	0.74 *	0.57 *
728	R	0.71 *	0.58 *
758	R	0.69 *	0.64 *
E 1616	R	0.71 *	0.70 *
G 550E	R	0.77 *	0.64 *
DK 18	S	0.72 *	0.73 *
DK 28	S	0.88 *	0.85 *
6073	S	0.79 *	0.74 *
HW 7217	S	0.53 *	0.74 *
HW 2194	S	0.82 *	0.82 *

[1]R and S represent resistant and susceptible nearly isogenic sorghum hybrids.
[2]LA and DWT represent percent leaf area and plant dry weight reductions.
* represent significance and nonsignificance at $P \leq 0.05$ by t-test.

Table 9. Mean of life stages (days) and fecundity (nymphs/female) of greenbug (GBE) on seedling sorghum hybrids.

Hybrid pairs[1]		PRLS[2]	TLS[2]	FEC[2]
708	R	9.73	36.36	43.45
		ns	ns	ns
DK 18	S	5.83	29.58	48.83
728	R	10.08	37.67	50.08
		ns	ns	ns
DK 28	S	9.18	37.18	53.27
758	R	4.18	27.09	28.82
		*	*	*
6073	S	7.75	35.42	47.92
E 1616	R	5.00	32.73	41.91
		*	ns	*
HW 7217	S	7.67	35.17	51.00
G 550E	R	4.83	30.50	39.17
		ns	ns	ns
HW 2194	S	5.36	28.36	41.55

[1]R and S represent resistant and susceptible nearly isogenic sorghum hybrids.
[2]PRLS=post-reproductive life span TLS=total life span and FEC=fecundity (nymphs/female).
* and ns represent significance and nonsignificance at $P \leq 0.05$ by paired t-test.

Table 10. Mean of life stages (days) and fecundity (nymphs/female) of greenbug (GBE) on resistant and susceptible seedling sorghum.[1]

Hybrids	PRLS[2]	TLS[2]	FEC[2]
Resistant			
728	10.08 a	37.67 a	50.08 a
708	9.73 a	36.36 ab	43.45 ab
E 1616	5.00 b	32.73 a	41.91 bc
G 550E	4.83 b	30.50 bc	39.17 b
758	4.18 b	27.09 c	28.82 c
Susceptible			
DK 28	9.18 a	37.18 a	53.27 a
6073	7.67 a	35.42 ab	47.92 a
HW 7217	7.67 a	35.17 ab	51.00 a
DK 18	5.83 a	29.58 bc	48.83 a
HW 2194	5.36 a	28.38 c	41.55 a

[1]Means with the same letter are not significantly different at $P \leq 0.05$ by LSD, when comparing resistant to resistant and susceptible to susceptible hybrids.
[2]PRLS, TLS and FEC represent post-reproductive life span, total life span (all in days) and fecundity (nymphs/female), respectively.

Table 11. Average number of greenbugs counted on five pair of seedling nearly isogenic sorghum hybrids.

Hybrid pairs[1]		Average number of greenbugs after 8 days
708	R	42.90
		ns
DK 18	S	43.60
728	R	48.75
		ns
DK 28	S	54.95
758	R	21.57
		*
6073	S	41.10
E 1616	R	30.85
		*
HW 7217	S	64.90
G 550	R	37.50
		*
HW 2194	S	52.55

[1]R and S represent resistant and susceptible nearly isogenic sorghum hybrids.

* and ns represent significance and nonsignificance at $P \leq 0.05$ by paired t-test.

Table 12. Average number of greenbugs counted on five pair of nearly isogenic sorghum hybrids.[1]

Hybrids	Average number of greenbugs after 8 days
Resistant	
728	48.75 a
708	42.90 ab
G 550E	37.50 bc
E 1616	30.85 bc
758	21.75 c
Susceptible	
HW 7217	64.90 a
DK 28	54.95 ab
HW 2194	52.55 ab
DK 18	43.60 b
6073	41.10 b

[1]Means with the same letter are not significantly different at $P \leq 0.05$ by LSD, when comparing resistant to resistant and sucseptible to susceptible hybrids.

Table 13. Greenbug response to nearly isogenic sorghum hybrids.[1]

Hybrid pairs[2]		Greenbug counts
708	R	1.30
		ns
DK 18	S	1.30
728	R	1.27
		ns
DK 28	S	1.38
758	R	1.10
		*
6073	S	1.27
E 1616	R	1.22
		*
HW 7217	S	1.41
G 550E	R	1.46
		ns
HW 2194	S	1.42

[1]Responses are greenbug counts converted into proportions of the initial number of greenbugs and then transformed by the arcsine transformation.
R and S represent resistant and susceptible nearly isogenic hybrids.
* and ns represent significance and nonsignificance at $P \leq 0.05$ by paired t-test.

Table 14. Plant height index (PHI) and dry weight index (DWI) as affected by greenbugs (GBE) for a period of 8 days.

Hybrid pairs[1]		PHI(%)	DWI(%)
708	R	2.48	1.02
		ns	ns
DK 18	S	3.16	1.26
728	R	1.69	0.76
		*	*
DK 28	S	3.02	1.43
758	R	2.96	0.54
		ns	*
6073	S	3.54	2.39
E 1616	R	3.25	1.67
		ns	ns
HW 7217	S	2.92	1.70
G 550E	R	2.27	1.21
		ns	ns
HW 2194	S	3.08	1.53

[1]R and S represent resistant and susceptible hybrids.
* and ns represent significance and nonsignificance at $P \leq 0.05$ by paired t-test.

Table 15. Plant height index (PHI) and dry weight index (DWT) for resistant and susceptible sorghum hybrids.[1]

Hybrids	PHI(%)	DWI(%)
Resistant		
E 1616	3.25 a	1.67 a
758	2.96 a	0.54 b
708	2.48 ab	1.02 ab
G 550E	2.27 ab	1.21 ab
728	1.69 b	0.76 b
Susceptible		
6073	3.54 a	2.39 a
DK 18	3.16 a	1.26 b
HW 2194	3.08 a	1.53 b
DK 28	3.02 a	1.43 b
HW 7217	2.92 a	1.70 b

[1]Means with the same letter are not signinificantly different at $P \leq 0.05$ by LSD, when comparing resistant to resistant and susceptible to susceptible hybrids.

PART II

FIELD EVALUATION OF ADVANCE GROWTH STAGE SORGHUM

HYBRIDS RELATIVE TO GREENBUG BIOTYPE E.

Abstract

The response to and damage caused by greenbug (GBE) on advance growth stage nearly isogenic sorghum hybrids was studied in the field.

The resistant member of nearly isogenic sorghum hybrid pair G 550E(R)/HW 2194(S) showed high levels of antibiosis and/or antixenosis. Under cage conditions significant differences were observed in fecundity between the members of this pair as more greenbugs were produced on the susceptible than the resistant member. When damage ratings were calculated, the susceptible member of each hybrid pair was significantly more damaged than the resistant member in four of the five comparisons. Under natural field conditions greenbug numbers on three pairs of nearly isogenic sorghum hybrids differed significantly. Larger greenbug numbers were present on the susceptible than the resistant member.

INTRODUCTION

The greenbug, *Schizaphis graminum* (Rondani) (Homoptera:Aphididae), has been a serious pest of wheat, *Triticum aestivum* L., since 1882 and of sorghum, *Sorghum bicolor* (L.) Moench, since 1968 (Kindler *et al*. 1984). Greenbugs can cause substantial yield losses in crops by the direct effects of feeding or as vectors of several plant viruses (Walgenbach *et al*., 1988). Greenbugs reproduce rapidly and mainly, if not entirely, by parthenogenesis (Mayo and Starks, 1972). When extremely abundant, greenbugs cause reduction in root and leaf development, the number of tillers is usually reduced, and the plant may be killed (Higgins and Brooks 1987). High rates of persistant systemic insecticides were initially relied on to control greenbugs in sorghum. These treatments were effective but at the same time had broad-spectrum toxicity and were environmentally disruptive (Young and Teetes 1977). Therefore, alternative approaches affording more economical control with less environmetal contamination were sought. One such approach was greenbug resistance in sorghum (Harvey and Hackerott, 1969. Wood, E.A Jr. 1971). Since then many sorghum hybrids resistant to greenbugs have been released. The purpose of this work was to screen recently released sorghum hybrids against greenbug biotype E in the field.

The objectives was:

Determine the response to and effect of greenbugs on advance growth stage sorghum hybrids in the field.

MATERIALS AND METHODS

Response to and Effect of Greenbug Biotype-E on Advance Growth Stage Sorghum Hybrids in the Field Under Cage Conditions

Field plots were established utilizing five nearly isogenic sorghum hybrid pairs to determine advance growth stage resistance against greenbug biotype E. The experiment was set up in a randomized complete block design with eight replications at Manhattan, Kansas. Each plot consisted of two rows x 9.14m. Greenbugs were placed on one plant per row. Clip-on-cages 1.5cm diameter were attached to the leaf by two clips to confine greenbugs. Plants were infested on two occasions. The first infestation was done when the sorghum plants were in the boot stage (i.e. head extended into flag leaf sheath) and the second was when the plants were at half bloom (Vanderlip, 1979). Two adult greenbugs were placed in each cage and cages attached to the underside of the lower leaf. Greenbug counts were made one week post-infestation. Damage ratings were made on a 0-10 scale based on the percent of the area within the cage with necrotic tissues. The greenbug counts for each hybrid were divided by the initial number of greenbugs infested to obtain fecundity. Fecundity and damage ratings were compared between the members of the nearly isogenic pairs by the use of a paired t-test and all means analyzed and separated by least significant difference (LSD) test.

The nearly isogenic sorghum hybrid pairs are as follow:

Resistant	Susceptible
708	DK 18
728	DK 28
758	6073
E 1616	HW 7217
G 550E	HW 2194

Greenbug Numbers on Advance Growth Stage Sorghum Plants in the Field Under Natural Condition

Three pairs of nearly isogenic sorghum hybrids used in the previous experiments were grown at the Kansas State University Experimental Field in Hays, Kansas to determine the response of a natural greenbug (GBE) infestation on advance growth stage sorghum plants. The three pairs were: 728(R)/DK 18(S), 728(R)/DK 28(S) and 758(R)/6073(S) which were selected solely on the basis of the availability of seeds. The experiment was conducted in a randomized complete block design with three replications. Each plot was a single row 5.18m long. Greenbug counts were conducted at 45, 52, 59 and 66 days after planting utilizing five plants per plot. The total number of greenbugs per hybrid was totaled for the four sampling periods indicated above. Total greenbug counts were divided by the number of plants from which greenbugs were counted (5), to get number of greenbugs/plant. Greenbug counts were compared by the use of paired t-tests between the members of the nearly isogenic pairs. Analysis of variance procedure and mean separation by least significant difference (LSD) were performed.

RESULTS AND DISCUSSION

Greenbug (GBE) Effect on Sorghum

Fecundity (nymphs/greenbug) and damage rating comparisons of nearly isogenic sorghum hybrid pairs grown in the field at Manhattan, Kansas are presented in Table 16.

The only pair which differed significantly in relation to fecundity was G 550E(R) and its nearly isogenic member HW 2194(S) (Table 16). These results agree with the results from the greenhouse experiments (Part I) as these members also differed significantly in those trials (see antibiosis part two). However, members of the two nearly isogenic hybrid pairs 758(R)/6073(S) and E 1616(R)/HW 2194(S) which differed significantly in the laboratory experiment, did not differ significantly in the field. Possible reasons for the non-significant differences may be attributed to the length of time the experiment was conducted. If the experimental duration was prolonged the resistant hybrids may have differed significantly from their respective susceptible counterpart. A second possibility is that the plants may not exhibit the same resistance mechanisms at different growth stages. The greenbug-induced damage was also rated and results presented in Table 16 for the nearly isogenic pairs. All resistant hybrids had significantly less damage than their susceptible counterparts except hybrid 728 (R) which did not differ significantly from DK 28(S). This could be attributed to the degree of resistance, in this case probably antibiosis, being less, see Table 16 under the column labeled fecundity, or the susceptible hybrid DK 28, its nearly isogenic pair, possessing a greater degree of antibiosis thereby resulting in no significant differences being

detected between the two. To get a better understanding of this relationship see Table 17 comparing the resistant hybrids among themselves and the susceptible hybrids to each other. As illustrated by this table hybrid 728 had the highest numerical damage rating of the resistant hybrids resulting from the second highest fecundity whereas DK 28 had the lowest damage rating of the susceptible hybrids. Resistant hybrids as a group did not differ significantly in fecundity among themselves nor did the susceptible hybrids as a group.

Greenbug numbers on Advance Growth Stage Sorghum Plants in the Field Under Natural Infestation

Results of this sampling are illustrated in Table 18 which presents the mean number of greenbugs per plant throughout the sampling period. It is evident from these results that there is some degree of antixenosis or antibiosis present in all hybrids deemed resistant as the number of greenbugs per plant was significantly less in each resistant member when compared to its susceptible member. When comparing the resistant hybrids as a group significant differences were observed for the number of greenbugs/plant but this was not the case for the susceptible hybrids (Table 19). In the greenhouse studies for antibiosis and antixenosis only one of the three resistant hybrids utilized in both trials, (758) had significantly reduced fecundity and less greenbugs per plant (in the choice test). The greenhouse studies were conducted on seedling plants for a much shorter time than the field trials. Thus, the lengthened time frame may have allowed these two mechanisms to better express themselves when resistant plants were compared to the

susceptibles. The resistant hybrids may also exhibit different mechanisms at different growth stages.

The difference between results from the Manhattan test and Hays test may be accounted for by:

(1) The length of infestation period at Hays was longer than at Manhattan.

(2) Greenbugs were caged at Manhattan whereas a natural field infestation occurred at Hays.

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Table 16. Fecundity (greenbugs/female) and damage rating to nearly isogenic sorghum hybrids by greenbug biotype E in the field.

Hybrid pairs[1]		Fecundity (greenbugs/female)	Damage rating
708	R	16.81	3.06
		ns	*
DK 18	S	16.63	4.21
728	R	17.16	2.78
		ns	ns
DK 28	S	13.61	3.38
758	R	14.97	2.69
		ns	*
6073	S	18.55	4.66
E 1616	R	15.64	2.34
		ns	*
HW 7217	S	17.97	4.72
G 550E	R	13.41	3.31
		*	*
HW 2194	S	18.91	4.00

[1]R and S represent resistant and susceptible nearly isogenic sorghum hybrids.
* significance and ns nonsignificance at $P \leq 0.05$ by paired t-test.

Table 17. Fecundity (greenbugs/female) and damage rating comparing resistant versus resistant and susceptible versus susceptible hybrids grown in the field.[1]

Hybrids	Fecundity (greenbugs/female)	Damage rating
Resistant		
708	16.81 a	3.06 a
728	17.16 a	2.78 a
E 1616	15.64 a	2.34 a
758	14.97 a	2.69 a
G 550E	13.41 a	3.31 a
Susceptible		
HW 7217	17.97 a	4.72 a
6073	18.55 a	4.66 a
DK 18	16.63 a	4.21 ab
HW 2194	18.91 a	4.00 ab
DK 28	13.61 a	3.38 b

[1]Means with the same letter are not significantly different at $P \leq 0.05$ by LSD, when comparing resistant to resistant and susceptible to susceptible hybrids.

Table 18. Mean number of greenbugs (GBE) under natural field infestation at Hays, KS.[1]

Hybrid pairs[2]		Greenbugs per plant
708	R	61.08
		*
DK 18	S	123.00
728	R	37.33
		*
DK 28	S	110.75
758	R	19.50
		*
6073	S	75.17

[1] * significance and ns nonsignificance at $P \leq 0.05$ by paired t-test.
[2] R and S represent resistant and susceptible nearly isogenic sorghum hybrids.

Table 19. Mean nummber of greenbugs (GBE) for resistant versus resistant and susceptible versus susceptible hybrids under a natural infestation at Hays, KS[1]

Hybrids	Greenbugs/plant
Resistant	
708	61.08 a
728	37.33 ab
758	19.50 b
Susceptible	
DK 18	123.00 a
DK 28	110.75 a
6073	75.17 a

[1]Means with the same letter are not significantly different at $P \leq 0.05$ by LSD, when comparing resistant to resistant and susceptible to susceptible hybrids.

SUMMARY AND CONLUSIONS

Laboratory evaluation of the nearly isogenic sorghum hybrids demonstrated that greenbug populations can damage and/or kill sorghum hybrids. The effect of greenbugs was more on the susceptible than the resistant hybrids as was observed by differences in both leaf area and plant dry weight in the nearly isogenic hybrid pairs E 1616(R)/HW 7217(S) and G 550E(R)/HW 2194(S). High levels of greenbug resistance were observed in hybrid E 1616(R) and G 550E(R) followed by hybrid 758(R) while the most susceptible hybrid was found to be DK 28. High levels of resistance in these hybrids may be attributed to relatively high degrees of antibiosis and/or antixenosis observed in the resistant member of these nearly isogenic pairs. This was observed when members of each nearly isogenic pair differed significantly in terms of antibiosis and antixenosis. The hybrid which demonstrated the highest level of antibiosis and antixenosis against greenbug biotype E was 758(R). Hybrid pair G 550E(R)/HW 2194(S) did not show antixenotic effects to greenbugs as members in this hybrid pair did not differ significantly. Hybrid pair 728(R)/DK 28(S), which did not display any of the mechanisms of resistance mentioned above, demonstrated high levels of tolerance. The resistant member of hybrid pair (758(R)/6073(S)) possessed some degree of tolerance as indicated by the differences in plant weight index between the members of the pair. Analysis of resistant hybrids determined that hybrid 728(R) had the highest levels of tolerance compared to the other resistant hybrids. The antibiotic and/or antixenotic effects observed in the resistant member of the nearly isogenic hybrid pair 758(R)/6073(S) was confirmed when greenbug feeding

behavior was studied. Greenbugs spent longer time feeding on the susceptible hybrid 6073 than they did on the resistant member. This isogenic pair, however, did not differ significantly when antibiosis (as measured by fecundity) was studied in cages for a week in the field. The same was true with the other isogenic pairs except G 550E(R)/HW 2194(S), which differed significantly between the members of the pair for all the parameters tested. Greenbug counts in the field over four sampling dates during the growing season differed significantly between the members of each pair for the three pairs (i.e. 708(R)/DK 18(S), 728(R)/DK 28(S) and 758(R)/6073(S)). The resistant nearly isogenic member in each pair had fewer greenbugs than its susceptible counterpart.

GREENBUG (SCHIZAPHIS GRAMINUM RONDANI)

BIOTYPE E INTERACTION WITH SORGHUM (SORGHUM BICOLOR L.)

HYBRIDS.

by

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Abstract

The effect of varying greenbug infestation levels, identification of mechanisms or components of resistance and feeding behavior of greenbug biotype E were studied on seedling, nearly isogenic sorghum hybrids in the laboratory. Similarly, an interactive effect of nearly isogenic sorghum hybrids and the relationship between greenbug numbers on advance growth stage sorghum in the field were determined.

Increased greenbug infestation levels affected sorghum seedlings by reducing leaf area and hence, plant dry weight. However, the effect was more on the susceptible than the resistant hybrids. The resistant member of nearly isogenic pairs which demonstrated relatively high levels of resistance include E 1616(R)/HW 7217(S), G 550E(R)/HW 2194(S) and 758(R)/6073(S). Antibiosis and/or antixenosis were the main components of resistance in these hybrids. The highest antibiosis and antixenosis levels were found in the resistant member of the nearly isogenic hybrid pair 758(R)/6073(S) and high levels of tolerance were observed in the resistant member of the nearly isogenic hybrid pair 728(R)/DK 28(S). The feeding behavior studies indicated that greenbug biotype E spent more time feeding on the susceptible hybrid 6073 than they did on the resistant member 758.

In the field experiment, the resistant member of the nearly isogenic sorghum pair G 550E(R)/HW 2194(S) demonstrated high levels of antibiosis and/or antixenosis as a larger number of greenbugs were found on the susceptible than the resistant member. Greenbug counts in the field over four sampling dates during the growing season differed significantly between the members of each pair for the three pairs (i.e.

708(R)/DK 18(S), 728(R)/DK 28(S) and 758(R)/6073(S)). The resistant nearly isogenic member in each pair had less greenbug counts than the susceptible counterpart.